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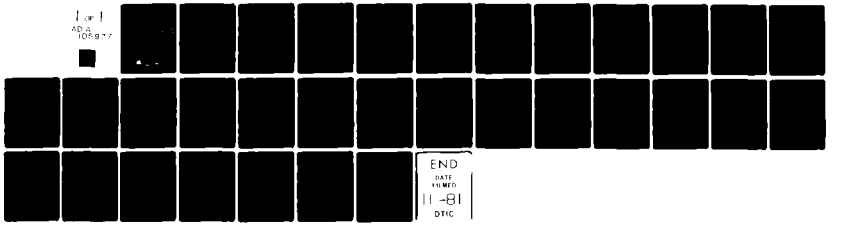

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INSTITUTE REPORT NO. 101

PRIMARY DERMAL IRRITATION POTENTIAL OF COMPONENTS OF THE M-258A-1 DECONTAMINATION KIT (Study 1)

JOHN T. FRUIN, DVM, PhD, LTC VC
and
MARTHA A. HANES, DVM, CPT VC

TOXICOLOGY GROUP,
DIVISION OF RESEARCH SUPPORT

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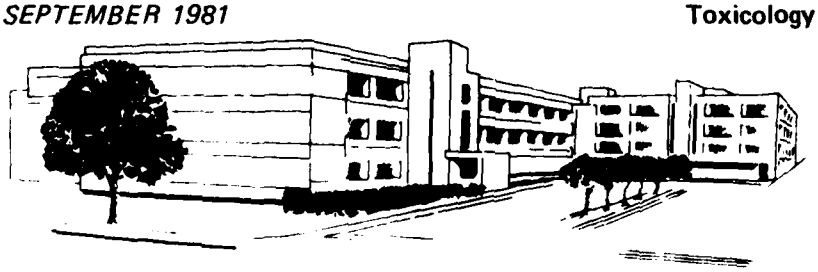

LETTERMAN ARMY INSTITUTE OF RESEARCH PRESIDIO OF SAN FRANCISCO CALIFORNIA 94129

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Toxicology Series 6

In conducting the research described in this report, the investigation adhered to the "Guide for the Care and Use of Laboratory Animals," as promulgated by the Committee on Revision of the Guide for Laboratory Animal Facilities and Care, Institute of Laboratory Animal Resources, National Research Council.

This material has been reviewed by Letterman Army Institute of Research and there is no objection to its presentation and/or publication. The opinions or assertions contained herein are the private views of the author(s) and are not to be construed as official or as reflecting the views of the Department of the Army or the Department of Defense. *(AR 360-5)*

John D. Marshall Jr 8 Sept 81
(Signature and date)

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M-258 Kit, Primary Dermal Irritation, Chemical Defense, Chemical Decontamination

20. ABSTRACT *(Continue on reverse side if necessary and identify by block number)*

The dermal irritation potential of components of the Prototype M-258A-1 decontamination kit was assessed by using the modified Draize test. The test called for approximately 0.5 g of material to be held in contact with the skin of rabbits for 24 hr. Both components, Decon I and Decon II, when applied separately and together caused severe erythema and necrosis after 24 hr. Further testing of these components is recommended to determine if they present a significant hazard.

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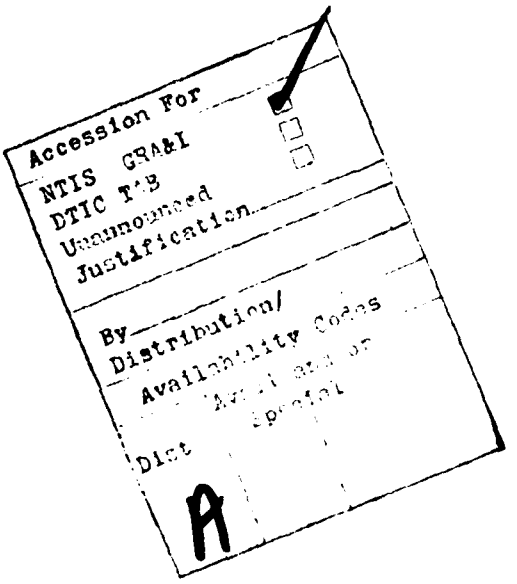
ABSTRACT

The dermal irritation potential of components of the Prototype M-258A-1 decontamination kit was assessed by using the modified Draize test. The test called for approximately 0.5 g of material to be held in contact with the skin of rabbits for 24 hours. Both components, Decon I and Decon II, when applied separately and together caused severe erythema and necrosis after 24 hours. Further testing of these components is recommended to determine if they present a significant hazard.



PREFACE

Primary Dermal Irritation Report

TESTING FACILITY: Letterman Army Institute of Research
Presidio of San Francisco, CA 94129

SPONSOR: Letterman Army Institute of Research
Presidio of San Francisco, CA 94129

PROJECT: Medical Defense Against Chemical Agents 612772.875.

GLP STUDY NUMBER: 81011

STUDY DIRECTOR: LTC (P) John T. Fruin, DVM, PhD, VC, Diplomate of American College of Veterinary Preventive Medicine

PRINCIPAL INVESTIGATOR: CPT Martha A. Hanes, DVM, VC

PATHOLOGY REPORTING: CPT George T. Makovec, DVM, VC
MAJ Glen E. Marrs Jr., DVM, MS, VC, Diplomate of American College of Veterinary Pathologists.

RAW DATA: A copy of the final report, study protocol, raw data, and standard operating procedures will be retained in the LAIR Archives.

TEST SUBSTANCES:

A. Decon I consists of a pad pre-wetted with hydroxyethane (ethanol) 72 $\pm$ 2%, phenol 10 $\pm$ 0.5%, sodium hydroxide 5 + 0.5%, ammonium hydroxide 0.2 $\pm$ 0.05% and water.

B. Decon II consists of a pad impregnated with a quantity of crystalline chloramine B and an equal quantity of liquid contained in breakable glass ampoules covered with nylon mesh. The liquid contains hydroxyethane (ethanol) 45 $\pm$ 2%, zinc chloride 5$\pm$0.5% and water. Just before dosing, the ampoules were broken and thus the chloramine B impregnated pad was saturated with liquid.

C. Decon I and Decon II combined

D. Control (dry 1 inch square cotton gauze pad)

WORK UNIT: 302 Studies on Potential Dermal Irritation of M-258A-1 Kit

PURPOSE: The purpose of this study was to determine the primary dermal irritation potential of the test substance listed above.

ACKNOWLEDGMENTS

The authors wish to thank LTC Kenneth Black MD, MC; SP5 Lance White; SP5 Leonard Sauers, BA; SP4 Thomas Kellner, BS; SP4 Larry Mullen, BS; PFC Evelyn Zimmerman; John Dacey; Carolyn Lewis, MS; for assistance in performing the research, and for advice in scoring the irritation reactions. The authors also wish to thank M. Mershon, VMD; LTC (P) E. Houston, PhD, MS; LTC R. Howarth, VMD, VC, of the U.S. Army Medical Research Institute of Chemical Defense, Aberdeen Proving Grounds, MD, for providing prototype M-258A-1 Decontamination Kits and background information.

Signatures of Principal Scientists Involved
In The Study

We, the undersigned, believe the study, GLP study number 81011 described in this report to be scientifically sound and the results and interpretation to be valid. The study was conducted to comply, to the best of our ability, with the Good Laboratory Practice Regulations for Nonclinical Laboratory Studies outlined by the Food and Drug Administration.

Martha A. Hanes, 21 Aug 81
MARTHA A. HANES, DVM / DATE
CPT, VC
Principal Investigator

John T Fruin 19 Aug 1981
JOHN T. FRUIN, DVM, PhD / DATE
LTC (P), VC
Study Director

DEPARTMENT OF THE ARMY
LETTERMAN ARMY INSTITUTE OF RESEARCH
PRESIDIO OF SAN FRANCISCO, CALIFORNIA 94129

SGRD-ULZ-QA

5 Aug 1981

MEMORANDUM FOR RECORD

SUBJECT: Report of GLP Compliance

I hereby certify that LAIR GLP study 81011 was a Primary Dermal Irritation Test study run concurrently and co-located with study 81007. Study records for 81011 were inspected during the inspection of 81007 on:

11 May 81

Findings were reported to the Study Director on 15 May 1981. This inspection is also included in the July 1981 report to management.

JOHN C. JOHNSON
CPT, MS
Quality Assurance Officer

TABLE OF CONTENTS

The urgent requirement for defensive measures and decontamination procedures was dramatically demonstrated to allied powers in the European Theater during World War I by the mass casualties resulting from the use of blister agents. Losses to the allies were high, because they lacked defensive capabilities. Probably more important they lacked sufficient offensive chemical warfare capabilities to deter the enemy from using chemical agents. The unclassified documentation of the use of antipersonnel chemical warfare agents since World War I has been difficult because the offensive use of chemical agents is a violation of treaties and creates negative world opinion. The clandestine use of offensive antipersonnel agents against guerrilla and poorly equipped forces has been reported in the popular press in recent years. The use of offensive chemical agents has also been suppressed because the major military powers possess chemical warfare capability and have not initiated chemical attacks for fear of similar retaliatory strikes. Current Eastern Block doctrine, although rarely if ever published or discussed publicly, is presumed to stress the use of munitions containing chemical agents integrated into conventional fire patterns (1). Thus there remains an urgent need for the United States and her allies to continue to pursue vigorously research and development efforts in the area of medical defense against chemical warfare agents. The rapid removal of chemical agents from the skin is an essential element in protecting personnel from chemical agents. The M-13 kit, a Fullers earth preparation, was developed before the M-258 kit as a means to remove chemical material by absorption (2,3). In addition to the physical removal of chemical agents, active chemical neutralization is desirable. Neutralization continues after protective gloves, mask and garments are in place. The M-258 decontamination kit, which is currently in the Army system, combines both physical removal and active chemical neutralization.

The original M-258 decontamination kit was type classified in the mid 1970s. It consisted of a plastic carrying case containing two plastic scrapers, a plastic container of Decon I solution, a plastic container of Decon II solution with a glass ampoule of chloramine B enclosed, and cotton gauze pads. The instructions contained on the plastic container directed the user to remove any suspected chemical agent with the plastic scrapers. The user was

instructed to open the Decon I container with the point of a nail which has its head imbedded in the plastic container lid. He was then instructed to saturate one of the cotton gauze pads and wipe exposed and contaminated area for one minute. The user was then instructed to break the glass ampoule inside the Decon II, shake well, open Decon II with the nail, saturate another cotton gauze pad and wipe the same area for 2 to 3 minutes (2).

The difficulty soldiers had in using the kit correctly , the caustic and irritating action of the chemical components, the abrasive action of wiping a limited area with a gauze pad for 4 minutes, and the potential for abrasion by broken glass from the Decon II container, were serious shortcomings of the M-258 kit (4). Nevertheless, the benefit/risk evaluation of the kit was favorable, but the need for continued research development efforts was recognized (Letter from US Armament Research and Development Command, DADAR-CLW-E, to US Army Training and Doctrine Command, ATCD-CF-S, Subject: Product Improvement proposal for Decontamination Kit M-258, dated 17 November 1977). Modifications already developed have made the kit much easier to use, reduced risk due to abrasion by broken glass, and reduced abrasion risk from the materials used to wipe the exposed skin (Contract Report ARCSL-CR-79046 to US Army Armament Research and Development Command for Contract DAAKII-78- C-0025, Subject: Feasibility of personnel Decontamination Kit, dated December 1979). The modified kit M-258A-1 consists of the plastic containers with six flex pouches. Three of the flex pouches contain pads saturated with Decon I solution. The remaining three pouches each contain pads impregnated with Chloramine B, and breakable glass vials of Decon II solution inside a nylon mesh bag designed to contain glass particles (5).

Deviation from standards

Rather than applying liquid test substance on gauze, liquid impregnated pads from the M-258A-1 decontamination kit were cut into roughly one inch squares. Decon I squares weighed about 0.3 g each, Decon II squares weighed about 0.6 g each. For Decon I dosing, two squares were applied one on top of the other. For Decon II dosing, one square was applied. For Decon I plus Decon II a smaller square (0.5 to 0.7 in) Decon II pad was sandwiched between two Decon I pads of similar size. Using these dosing procedures, we anticipated on effective dose level of 0.5 g test substance per site.

Chemical analyses were not conducted except for measuring pH. The pH of Decon I was 10.7-10.8, Decon II was 6.5-6.6, and combined Decon I and II was 10.6-10.7. Chemical composition was considered to be that printed on the outer container for the prototype M-258A-1 decontamination kit (Tables 1 and 2). Compound stability and purity are unknown.

TABLE 1 (6)

CHEMICAL ANALYSIS OF DECON I
(pH = 10.7 - 10.8)

Component	ETOH	H_2O	Phenol	NaOH	NH_4OH
%	72%±2%	q.s.	10±0.5%	5.0±0.5%	0.2±0.05%
Name	ethanol	water	phenol	sodium hydroxide	ammonium hydroxide
Molecular Structure	C_2H_6O	H_2O	C_6H_6O	NaOH	NH_4OH
Molecular Weight	46.07	18.016	94.12	40.01	35.036

TABLE 2 (6)

CHEMICAL ANALYSIS OF DECON II
(pH = 6.5 - 6.6)

	*LIQUID PORTION			*SOLID PORTION
Component	ETOH	H_2O	$ZnCl_2$	Chloramine B
%	45±2%	50±2.5%	5±0.5%	100%
Name	ethanol	water	zinc chloride	Chloramine B (N-Chlorobenzene-sulfamido-sodium)
Molecular Structure	C_2H_6O	H_2O	$ZnCl_2$	$C_6H_5Cl\ NNaO_2S$
Molecular Weight	46.07	18.016	136.29	213.64

* Equal quantities of liquid and solid are mixed to form Decon II.

Objective of Study

The objective of this study was to determine the primary dermal irritation potential of the components of the Prototype M-258A-1 decontamination kit.

METHODS

Historical Listing of Study Events

16 April 1981	Rabbits arrived, they were weighed, tattooed, sexed, and quarantined for two weeks.
23 April 1981	Animals were weighed.
30 April 1981	Animals were released from quarantine, close clipped, and marked.
1 May 1981	Animals were weighed, randomized into two specific groups and sites for exposure randomized.
4 May 1981	Animals were dosed according to SOP-OP-STX-34, except as stated in deviations from standards. Animals were observed daily, only significant or abnormal observations were recorded.
5 May 1981	Bandages were removed and the 24-hour score was taken. One animal was sacrificed for histopathology of exposure sites.
7 May 1981	Animals were weighed and the 72-hour score was taken. One animal was sacrificed for histopathology of exposure sites.
11 May 1981	The 7-day score was taken.
14 May 1981	The animals were weighed.
18 May 1981	The 14-day score was taken. Animals remained on study, as not all of irreversible damage had healed.
22 May 1981	A 18-day score was taken for animals still on study.
26 May 1981	A 22-day score was taken, all animals still on study were released.

Animal Data

Animal: New Zealand Rabbits

Sex: Male

Source: Elkhorn Rabbitry

Pre-test Conditioning:

A. Quarantine from 16 April 1981 to 30 April 1981.

B. Animals were close-clipped and test areas marked.

Animal Identification: Right ear tattooed, SOP-OP-ARG-1.

Method of Randomization: Manual, Latin Square, SOP-OP-STX-34.

Number of Animals on test: 8 animals - each animal had 4 test sites and received each of the three test substances and a control patch.

Age of animals at start of study: young adults

Body Weight Range: 2.5 - 3.5 kg

Condition of animals at start of study: normal

Environmental Conditions

Caging: Number/cage = 1; Type cage used = stainless steel, wire mesh bottom, battery type, no bedding, automatic flushing.

Diet: Purina Certified Rabbit Chow #5322 approximately 110 g per day supplemented with about 45 g of fresh carrots.

Water: Central line to cage battery with automatic lick dispensers.

Temperature: 69 $\pm$ 5 F (21 $\pm$ 3 C)

Relative Humidity: about 60 $\pm$ 10%

Photoperiod: 0530 - 2000 hr/day (14 1/2 hr of light).

Dosing Levels

A. Approximately 0.5 g Decon I

B. Approximately 0.5 g Decon II

C. Approximately 0.25 g of Decon I and 0.25 g Decon II.

D. Control (nothing was applied)

Dosing Procedures

Method and frequency of administration were specified by Department of the Army (2) and Environmental Protection Agency (7). The backs of the animals were close-clipped and divided into quadrants designated I,II,III,and IV starting in the right shoulder area and proceeding clockwise. Areas I and IV were intact on all animals, and areas II and III were abraded by making two perpendicular scratches in the stratum corneum of the skin about 1 1/2 inch long by using an escarifer. The four application sites were about 10 cm apart. A standard latin square table was used to randomize the test sites (SOP-OP-STX-34).

The test substance impregnated pads were taped over the test sites. A plastic strip held on by elastic tape was placed around the animal to retard evaporation and to insure skin contact by the test substance. The test substance was in contact with the skin for 24 hours. At the end of the exposure period, the wrapping was removed, the skin wiped if material was adherent, and the areas were scored (8).

RESULTS

Scoring

Originally eight animals were exposed to the chemicals. Animals F8100025 and F8100017 were randomly chosen for histologic investigation of depth of damage at 24 and 72 hours, respectively. The pathology reports appear in Appendix A.

Seven animals were scored at 24 and 72 hr, 7,14,18, and 22 days for edema and erythema (Table 3). The primary skin irritation test data are summarized in Appendix B. Abraded (sites II and III) and intact areas (site I and IV) were graded separately as well as together. The scores obtained were used for a basis for categorization. Primary irritation potential values were calculated from the 24-and 72-hr scores.

TABLE 3

EVALUATION OF SKIN REACTIONS (9)

Erythema and Eschar Formation	
No erythema	0
Very shight erythema (barely perceptible)	1
Well-defined erythema	2
Moderate-to-severe erythema	3
Severe erythema (beet redness) to slight eschar formation (injurious in depth)	4
Possible total erythema score	4*
Edema Formation	
No edema	0
Very slight edema (barely perceptible)	1
Slight edema (edges of area well defined by definite raising)	2
Moderate edema (edges raised approximately 1 mm)	3
Severe edema (raised more than 1 mm and extending beyond area of exposure)	4
Possible total edema score	4*
Possible total score for primary irritation	8

*Any skin reaction more serious than severe edema, vesiculation, ulceration, or necrosis places the chemical in Category IV.

Compounds producing combined averages (intact and abraded scores) of 2 or less are considered mildly irritating (Category I), whereas those with indexes from 2 to 5 are moderate irritants (Categories II and III).

Category IV irritants are compounds producing moderate to severe primary irritation of intact skin and skin surrounding an abrasion. In addition these compounds produce necrosis, vesiculation, ulceration and/or eschars. (Category assignment and interpretation, personnel communication, AH McCreesh, 1980.)

Table 4 demonstrates the primary irritation indexes for the exposed areas.

TABLE 4

PRIMARY DERMAL IRRITATION INDEX FOR M-258A-1 DECONTAMINATION KITS

Chemical	Intact Score	Abraded Score	Combined Score	Category
Decon I	5.7	7.5	6.7	IV
Decon II	7.8	7.4	7.6	IV
Decon I+II	5.6	5.8	5.7	IV
Control	0.0	0.0	0.0	I

Clinical Observations

When we removed the bandages, we observed wet burns (Decon I) or dry black areas (Decon II) of burned skin under the entire areas of application. The combined sites of Decon I and II were lightly tinted blue-green at the fringes by the zinc-ammonia complex.

Animals exposed to test substances were anorexic and were extremely sensitive over the exposed areas. All animals exhibited scarring after loss of the scabs, which occurred by post-exposure day 18. Animals were restrained by hand for application of test substance and were not kept in restrainers for the initial period. The animals did not damage bandages or selfmutilate during the course of the study. Two animals exhibited post-exposure secondary infection on application sites. The infected animals were not treated. The infected sites appeared to clear in 3 to 4 days.

DISCUSSION

Decon I and Decon II showed severe irritation potential.

Reports that solutions of Decon I and Decon II when combined

neutralized each other were not confirmed by this study. They yielded slightly lower combined and abraded scores for these sites. From the clinical point of view, the lower scores for Decon I+II combined are purely academic. Severe damage and burning was produced by the Decontamination Kit constituents.

The interpretation for Category IV is that the compound should be resubmitted "in the form and the intended use concentrations" so that irritation potential can be reevaluated.

CONCLUSION

The prototype M-258A-1 kit contains potentially severe irritants.

RECOMMENDATIONS

It is recommended that the following studies be conducted to demonstrate the irritation potential of the chemicals when used as directed, or as they would be utilized in the field.

Six tests are suggested:

Tests 1 and 2 would include studies where the kit constituents would be used as directed on the plastic container; Tests 3 and 4 would include studies where freshly prepared chemicals contained in the kit would be applied in amounts needed to wet 1-inch square of rabbit skin; Tests 5 and 6 would include studies where physiological saline would be used to wipe the skin of rabbits for 1,3, and 4 minutes to simulate the directed use of the M-258A-1 Kit. Each pair of tests would include occluded test sites (Tests 1,3,5) and non-occluded test sites (Tests 2,4,6).

Occluded is intended to mean the use of a plastic wrap and adhesive to retard evaporation and assure juxtaposition of the chemical and skin. Non-occluded means the total absence of tape, gauze and any wrapping materials.

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Pathology Reports

APPENDIX A

APPENDIX A-1

LAIR 30524

A. Rabbit, New Zealand White, male, 2 months.

B. ID: F8100025.

C. Dr. Hanes, TL10, Decontamination - GLP #81011.

D. History: This animal was exposed to skin decontamination solutions from the M-258A-1 Decontamination kit. The dorsal thorax and lumbar areas were shaved and the solutions applied as follows:

Quadrant I - Decon II, normal skin (slide 1).

Quadrant II - Decon I & II, abraded skin (slide 2).

Quadrant III - Control, abraded skin (slide 3).

Quadrant IV - Decon I, normal skin (slide 4).

After 24 hours exposure time the rabbit was anesthetized with sodium pentobarbital, then exsanguinated by cutting the axillary arteries. Skin sections were harvested from each quadrant and fixed in 10% buffered neutral formalin. The skin sections were submitted to Pathology Services for histopathologic examination.

E. Gross findings: See history.

F. Morphologic findings:

Slide 1 - Skin: The skin overlying a narrow band of cutaneous muscle is almost devoid of epidermis. The surface is covered by a thin layer of flaking, fragmenting basophilic material with a few underlying remnants of epidermis at each end. Most of the epidermis is comprised of small cells with dense, eosinophilic cytoplasm and pyknotic or fragmenting nuclei. Several inflammatory cell foci, primarily heterophils with cell debris, underlie the epidermis or basophilic covering material. The entire epidermis is expanded and hypercellar with wide separation of collagen bundles. All structures in the upper 20% of the dermis including adnexia, are deeply eosinophilic and smudgy or granular. All the nuclei are pyknotic or fragmented. The eosinophilic zone is bounded by a prominent band of degenerating heterophils and debris. The irregularly straining and occasionally fragmenting collagen bands in the remaining dermis are widely separated and contain a prominent diffuse inflammatory cell infiltrate that is primarily heterophils with some lymphocytes, plasma cells and large mononuclear cells. There

are many variable sized irregularly shaped foci of lacy eosinophilic fibrillar material that often contains some free blood and are occasionally associated with small fragmenting vessels distributed throughout the dermis. Many of the spindle shaped cells throughout the dermis are rounded and occasionally have pyknotic nuclei.

DX: Necrosis, acute, diffuse, severe, epidermis, skin, rabbit.

DX: Degeneration acute, diffuse, dermis, skin.

Slide 2 - Skin: Essentially the same as slide 1 but the prominent band of inflammatory cell infiltrate is in the deep dermis, adjacent to but not involving the cutaneous muscle.

DX: Necrosis, acute, diffuse, severe, epidermis, skin.

DX: Degeneration, acute, diffuse, severe, dermis, skin.

Slide 3 - Skin: No lesions recognized (NLR), epidermis and dermis, skin.

Slide 4 - Skin: Essentially the same as slide 2.

DX: Necrosis, acute, diffuse, severe, epidermis, skin.

DX: Degeneration, acute, diffuse, severe, dermis, skin.

G. Morphologic diagnoses:

1. Necrosis, acute, diffuse, severe, epidermis, skin, rabbit- Decon I, Decon I & II, Decon II.

2. Degeneration, acute, diffuse, severe, dermis, skin - Decon I, Decon I & II, Decon II.

3. no recognized lesions, skin - Control.

H. Syndrome diagnosis: N/A.

I. Comment: The damage to the epidermis and upper dermis caused by Decon I, Decon I & II, and Decon II is probably due to direct caustic effect of the substances at high concentrations. The more subtle, but

still severe, effect in deeper dermis of skin sections exposed to Decon I, Decon I & II, and Decon II is probably a combination of direct toxic damage and hypoxia associated with vascular damage.

GEORGE T. MAKOVEC, DVM
CPT, VC
Pathology Services Group
Division of Research Support

GLEN E. MARRS, JR., DVM, MS
Diplomate of ACVP
MAJ, VC
Pathology Services Group
Division of Research Support

APPENDIX A-2

LAIR 30527

A. Animal Data: Rabbit, New Zealand White, male.

B. ID: F8100017.

C. Contributor: Dr. Hanes, TL10, GLP #81011.

D. History: This animal was exposed to skin decontamination solutions from the M-258A-1 Decontamination kit. The dorsal thorax and lumbar areas were shaved and the solutions applied as follows:

Quadrant I - Decon I & II, normal skin (slide 1).

Quadrant II - Control, abraded skin (slide 2).

Quadrant III - Decon I, abraded skin (slide 3).

Quadrant IV - Decon II, normal skin (slide 4).

After 72 hours exposure time the rabbit was anesthetized with sodium pentobarbital, then exsanguinated by cutting the axillary arteries. Skin sections were harvested from each quadrant and fixed in 10% buffered neutral formalin. The skin sections were submitted to Pathology Services for histopathologic examination.

E. Gross findings: See history.

F. Morphologic findings:

Slide 1 - Skin: The skin is covered by a heavily keratinized epithelium that is intact except for two small foci in which the remaining epithelial cells have dense eosinophilic cytoplasm and pyknotic nuclei. These foci contain a prominent inflammatory cell infiltrate, primarily heterophils, and cellular debris. In the upper 15% of the dermis, even under the intact epidermis, the collagen bundles are lightly eosinophilic, fragmented and granular. This portion of the dermis contains a moderate inflammatory cell infiltrate, comprised primarily of heterophils, with a few lymphocytes, plasma cells and mononuclear cells. Pavementing by heterophils is observed in a few small vessels within the altered area. There are a few small aggregates of lymphocytes and plasma cells with an occasional heterophil in the connective tissue sheaths of a few hair follicles deep in the dermis.

DX: Necrosis, acute, focal, severe, epidermis, skin, rabbit.

DX: Degeneration, acute, diffuse, mild-moderate, dermis, skin.

DX: Folliculitis, subacute, multifocal, minimal, hair follicles, skin.

Slide 2 - Skin: There are a few small aggregates of plasma cells and lymphocytes with an occasional heterophil in the connective tissue sheaths of a few hair follicles deep in the dermis.

DX: Folliculitis, subacute, multifocal, minimal, hair follicles, skin.

Slide 3 - Skin: Essentially the same as slide 1, but the epidermis is intact.

DX: Degeneration, acute, diffuse, mild to moderate, dermis, skin.

DX: Folliculitis, subacute, multifocal, minimal, hair follicles, skin.

Slide 4 - Skin: The entire skin surface is covered by a thick layer of densely eosinophilic material containing much cell debris. This material is almost completely separated from the dermis by a thin layer of epidermis. The epidermis is generally only one or two cells thick except in follicular areas where the epithelium piles and an occasional mitotic figure is present. The dermis adjacent to the epidermis contains a small amount of granular debris and inflammatory cell infiltrate that is comprised primarily of large mononuclear cells with granular cytoplasm and an occassional heterophil, lympocyte or plasma cell. There are a few small aggregates of plasma cells and lymphocytes with an occasional heterophil in the connective tissue sheaths of a few hair follicles deep in the dermis.

DX: Hyperplasia, diffuse, moderate, epidermis, skin.

DX: Degeneration, subacute, diffuse, minimal to mild, dermis, skin.

DX: Folliculitis, subacute, multifocal, minimal, hair follicles, skin.

G. Morphologic diagnoses:

1. Necrosis, acute, diffuse, severe, epidermis, skin, rabbit-Decon I & II.

2. Hyperplasia, diffuse, moderate, epidermis, skin - Decon I.

3. Degeneration, acute to subacute,, diffuse, mild to moderate, dermis, skin - Decon I, Decon I & II.

4. Degeneration, subacute, diffuse, minimal to mild, dermis, skin - Decon II.

5. Folliculitis, subacute, multifocal, minimal, hair follicles, skin Decon I & II, Decon I, Decon II, control.

H. Syndrome diagnosis: N/A.

I. Comment: The damage to the epidermis and upper dermis caused by Decon I, Decon I & II, and Decon II is probably due to both direct and indirect toxic effect of the substances tested. The regeneration of the epithelium beneath the thick layer of debris on the skin exposed to Decon II suggests that the necrotic surface material may serve as a protective barrier. The minimal folliculitis in all the skin sections is considered to be an incidental finding and not related to exposure to the compounds tested.

for

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Summary of Primary Skin Irritation Test Data

APPENDIX B

APPENDIX B-1

Summary of Primary Skin Irritation Test Data

GLP Study No. 81011

Chemical Name	Conc	Solvent	Amt Applied	Form
M-258A-1 Decon I	*	NA	0.5 g	A

Date of Application 4 May 1981

Principal Investigator CPT HANES

*2 layers of 1" square section of towelett.

Irritation Scores

		Intact Skin Sites				Abraded Skin Sites				
Rabbit No.	Site	Erythema 24 hr	Erythema 72 hr	Edema 24 hr	Edema 72 hr	Site	Erythema 24 hr	Erythema 72 hr	Edema 24 hr	Edema 72 hr
F8100015	I	2	4	3	3					
F8100016	IV	2	4	1	1					
F8100017						III	4	4	4	2
F8100022						II	4	4	4	4
F8100023	I	4	4	2	4					
F8100025	IV	*	*	*	*					
F8100026						III	4	4	4	4
F8100032						II	4	4	2	4
Total:		a 8	b 12	a 6	b 8		a 16	b 16	a 14	b 14
		a+b 20		a+b 14			a+b 32		a+b 28	

C^I + 34

C^A + 60

Intact Score = C^I / 2xNo. of Sites on test 34/(3x2) = 5.7

Abraded Score = C^A / 2xNo. of Sites on test 60/(4x2) = 7.5

Total Score = $\frac{C^I + C^A}{2}$ x No. of Sites on test (34+60)/(7x2) = 6.7

Primary Skin Irritation Index Category IV

Remarks: *F8100025 was randomly chosen for sacrifice at 24 hr for histological study. F8100017 was chosen randomly for sacrifice at 72 hr for histological study.

APPENDIX B-2

Summary of Primary Skin Irritation Test Data

GLP Study No. 81011	Chemical Name	Conc	Solvent	Amt Applied	Code
Date of Application 4 May 1981	M-258A-1 Decon II	*	NA	~0.5 g	B

Principal Investigator CPT HANES

* 1 layer of 1x1" section of towelette.

Irritation Scores

		Intact Skin Sites				Abraded Skin Sites				
Rabbit No.	Site	Erythema 24 hr	Erythema 72 hr	Edema 24 hr	Edema 72 hr	Site	Erythema 24 hr	Erythema 72 hr	Edema 24 hr	Edema 72 hr
F8100015						II	4	4	4	4
F8100016	I	4	4	4	3					
F8100017	IV	4	4	4	4					
F8100022						III	4	4	4	4
F8100023						II	4	4	3	4
F8100025	I	*	*	*	*					
F8100026	IV	4	4	4	4					
F8100032						III	1	4	3	4
Total:		a 12	b 12	a 12	b 11		a 13	b 16	a 14	b 16
		a+b 24		a+b 23			a+b 29		a+b 30	
		C^I + 47					C^A + 59			

Intact Score = C^I / 2xNo. of Sites on test 47/(3x2) = 7.8

Abraded Score = C^A / 2xNo. of Sites on test 59/(4x2) = 7.4

Total Score = $\frac{C^I + C^A}{2 \times No. \text{ of Sites on test}}$ (47+59)/(7x2) = 7.6

Primary Skin Irritation Index Category IV

Remarks: *F8100025 was chosen randomly for histological section after 24 hr.

F8100017 was chosen randomly for histological section after 72 hr.

APPENDIX B-3

Summary of Primary Skin Irritation Test Data

GLP Study No. 81011

Date of Application 4 May 1981

Principal Investigator CPT HANES

Chemical Name	Conc	Solvent	Amt Applied	Site
M-258A-1 Decon I & II	*	NA	~0.5 g	C

*1/2" x 1" of Decon II and 1x1" of Decon I

Irritation Scores

		Intact Skin Sites					Abraded Skin Sites			
Rabbit No.	Site	Erythema 24 hr	Erythema 72 hr	Edema 24 hr	Edema 72 hr	Site	Erythema 24 hr	Erythema 72 hr	Edema 24 hr	Edema 72 hr
F8100015						III	0	4	2	3
F8100016						II	4	4	4	2
F8100017	I	0	4	2	3					
F8100022	IV	0	4	2	4					
F8100023						III	1	4	4	3
F8100025						II	*	*	*	*
F8100026	I	1	4	4	4					
F8100032	IV	1	4	4	4					
Total:		a 2	b 16	a 12	b 15		a 5	b 12	a 10	b 8
		a+b 18		a+b 27			a+b 17		a+b 18	
		C^I = 45					C^A = 35			

Intact Score = C^I / 2xNo. of Sites on test 45/(4x2) = 5.6

Abraded Score = C^A / 2xNo. of Sites on test 35/(3x2) = 5.8

Total Score = $\frac{C^I + C^A}{2 \times \text{No. of Sites on test}}$ (45+35)/(7x2) = 5.7

Primary Skin Irritation Index Category IV

Remarks: *F8100025 was chosen randomly for histological section after 24 hr.

F8100017 was chosen randomly for histological section after 72 hr.

APPENDIX B-4

Summary of Primary Skin Irritation Test Data

GLP Study No. 81011

Date of Application 4 May 1981

Principal Investigator CPT HANES

Chemical Name	Conc	Solvent	Amt Applied	Code
M-258A-1 Control	NA	NA	NA	D

Irritation Scores

		Intact Skin Sites					Abraded Skin Sites				
Rabbit No.	Site	Erythema 24 hr	Erythema 72 hr	Edema 24 hr	Edema 72 hr	Site	Erythema 24 hr	Erythema 72 hr	Edema 24 hr	Edema 72 hr
F8100015	IV	0	0	0	0					
F8100016						III	0	0	0	0
F8100017						II	0	0	0	0
F8100022	I	0	0	0	0					
F8100023	IV	0	0	0	0					
F8100025						III	*	*	*	*
F8100026						II	0	0	0	0
F8100032	I	0	0	0	0					
Total:		a 0	b 0	a 0	b 0		a 0	b 0	a 0	b 0
		a+b 0		a+b 0			a+b 0		a+b 0	
		C^I + 0					C^A + 0			

Intact Score = C^I / 2xNo. of Sites on test 0/(4x2) = 0

Abraded Score = C^A / 2xNo. of Sites on test 0/(3x2) = 0

Total Score = $\frac{C^I + C^A}{2 \times \text{No. of Sites on test}}$ (0+0)/(7x2) = 0

Primary Skin Irritation Index Category I

Remarks: F8100025 was chosen randomly for histological section at 24 hr.

F8100017 was chosen randomly for histological section at 72 hr after dosing.

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